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(54) **DETECTION OF CYP3A4 AND CYP2C9 POLYMORPHISMS**

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(57) **ABSTRACT**

The invention provides oligonucleotide primer pairs, sequence determination oligonucleotides, and kits for amplification and detection of novel single nucleotide polymorphisms in the 5' flanking regions of the CYP3A4 and CYP2C9 genes.

FIGURE 1

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   1 CTGCAGTGAC CACTGCCCCA TCATTGCTGG CTGAGGTGGT TGGGGTCCAT CTGGCTATCT
  61 GGGCAGCTGT TCTCTTCTCT CCTTTCTCTC CTGTTTCCAG ACATGCAGTA TTTCCAGAGA
 121 GAAGGGGCCA CTCTTTGGCA AAGAACCTGT CTAACTTGCT ATCTATGGCA GGACCTTTGA
 181 AGGGTTCACA GGAAGCAGCA CAAATTGATA CTATTCCACC AAGCCATCAG CTCCATCTCA
 241 TCCATGCCCT GTCTCTCCTT TAGGGGTCCC CTTGCCAACA GAATCACAGA GGACCAGCCT
 301 GAAAGTGCAG AGACAGCAGC TGAGGCACAG CCAAGAGCTC TGGCTGTATT AATGACCTAA
 361 GAAGTCACCA GAAAGTCAGA AGGATGCATA GCAGAGGCCC AGCAATCTCA GCTAAGTCAA
 421 CTCCACCAGC CTTTCTAGTT GCCCACTGTG TGTACAGCAC SCTGGTAGGG ACCAGAGCCA
 481 TGACAGGGAA TAAGACTAGA CTATGCCCTT GAGGAGCTCA CCTCTGTTCA GGGAAACAGG
 541 CGTGGAAACA CAATGGTGGT AAAGAGGAAA GAGGACAATA GGATTGCATG AAGGGGATGG
 601 AAAGTGCCCA GGGGAGGAAA TGGTTACATC TGTGTGAGGA GTTTGGTGAG GAAAGACTCT
 661 AAGAGAAGGC TCTGTCTGTC TGGGTTTGGA AGGATGTGTA GGAGTCTTCT AGGGGGCACA
 721 GGCACACTCC AGGCATAGGT AAAGATCTGT AGGTGTGGCT TGTTGGGATG AATTTCAAGT
 781 ATTTTGGAAT GAGGACAGCC ATAGAGACAA GGGCARGAGA GAGGCGATTT AATAGATTTT
 841 ATGCCAATGG CTCCACTTGA GTTTCTGATA AGAACCCAGA ACCCTTGGAC TCCCCAGTAA
 901 CATTGATTGA GTTGTTTATG ATACCTCATA GAATATGAAC TCAAAGGAGG TCAGTGAGTG
 961 GTGTGTGTGT GATTCTTTGC CAACTTCCAA GGTGGAGAAG CCTCTTCCAA CTGCAGGCAG
1021 AGCACAGGTG GCCCTGCTAC TGGCTGCAGC TCCAGCCCTG CCTCCTTCTC TAGCATATAA
1081 ACAATCCAAC AGCCTCACTG AATCACTGCT GTGCAGGGCA GGAAAGCTCC ATGCACATAG
1141 CCCAGCAAAG AGCAACACAG AGCTGAAAGG AAGACTCAGA GGAGAGAGAT AAGTAAGGAA
1201 AGTAGTGATG GCTCTCATCC CAGACTTGGC CATGGAAACC TGGCTTCTCC TGGCTGTCAG
1261 CCTGGTGCTC CTCTATCTGT GAGTAACTGT TCAGGCTCCT CTTCTCTGTT TCTTGGACTT
1321 GGGGTCGTAA TCAGGCCTCT CTTTT
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FIGURE 2

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   1 GATCTCAGAT ATCCCTTCTA TCTACACATT ATCTATAATT CTTTCTTTCT GTAAACTGAA
  61 AGGTCCTAGA AGGAGCCGCA GCTCAGCAGG AGAGAGGAGG AGCTGAGCTG GGACCCCTAC
 121 CTCCTGAGGA ATGAAATGAT TATTATAAAG ACAGCAACCG AGCTTATTTT ACCCAAAATA
 181 AGGTAGTATA TTTCTGTTAG AGTTTAGAGT TTCATGAGTC AGGGACCAAG TTATTGCTTT
 241 TCTTTGCCCT GTATAAAGGC TTCTCCAAGG CCTTTGACTT ACCTAAGTAC TAAATGTTAT
 301 AAAACCAAAC TCTTCTGACC TCTCAATCTA GTCAACTGGG GCTGTAATTA TTAATGAAAT
 361 TAATGTTTAT TTTGAAAATA ATTTACTAGA CTGAATTACG AAATCCTGAA TCATTGTACA
 421 CTATCAGTAA ATATTGGTGG ACCCAACTGA ACTGAATGTT TTGCTTGAAA TGAAACCTTT
 481 GAGATGCAGG GCTTATGGGT TCTAGTCCCA GCTCTAGCAC TAGCAGACAG CATGTTCTTG
 541 GCTAAGATAC TGAATCTTCA AGGCTCAGCT TCCTCATTCC GGAAATGGGT CAATTTTATT
 601 GTAAGCAGAG GTAATTGAGA GATTCAAAAG GGACATGAGG TGTAACAATT CTCTGTAAAT
 661 TGTTAGAATC CCTGTTAAAA ATGACCAGTA AAGCTTTGTG CAACTGTGTC TTGACATAAC
 721 TTTATTTTTC TTAATAAAAG AAATGGAAAT AACCTCACTA GGGAATTTAG AACAAATATG
 781 ATGATATCTT TAAAGAAAAT GGCTTTGCAC AAGTATTGAC ATTAATGATC TAGTAAAGTG
 841 TATCTTTCTA GTTGTATTTA GATCCTCAAC TCAGTATGTC AGCTCCTGTT AAGGTCTATA
 901 CATTGTGGTG GTTCTGTGCT GTGGGTCCAT TTAGTGATTT CCCTACCTCC CATCTTYTAT
 961 TGCATCCACA ACTGTGGTTC TGTCCATAAT TTCCTTTGCT TTCTGTGCAT TATTACATCA
1021 TATCTGAAAA TGAGAAACCA AAAACAATRG AAAGCAGCCA TGTCTGGAGG TGACTGGGGG
1081 GTCGAGAAGC CCTAGTTTCT CAAACCCTTA GCACCAAATT TTTCCCTCAG TTACACTGAG
1141 CGTTTCACTT CTGCAGTGAT GGARAAGGGA GATCCCTTAT TTCTTCTCAT GAGCATCTCT
1201 GGTGCTGTTT CCCTTAGAGA CAAATAAGGG GTTCTATTTA ATGTGAAGCC TGTTTTATGA
1261 ACAGAATAAA TGTGGTGTAT ATTCAGAATA ACTAATGTTT GGAAGTTGTT TTATTTTTGC
1321 TAAAAATTGT TCTCAAGGCA GCTCTGGTGT AAGAGATAAT ACACCACGAT GGGCATCAGA
1381 AGACCTCAGC TCAAATCCCA GTTCTGCCAG CTATGAGCTG TGTGGCACCA ACAGGTGTCC
1441 TGTTCTCCCA GGGTCTCCCT TTTCCCATTT GAAAAATAAA AAATAACAAT TCCTGCCTTC
1501 AGGAATTTTT TTTAGGGGGT TTAATKGTAA AGGTGTTTAT ATCTGCTAAG GTAATTTACT
1561 TGATATATGT TTGGTTATTT AAGATATATG AGTTATGTTA GCTATTTCAT GTTTAGGCTG
1621 CTGTATTTTT AGTAGGCTAT ATTAAATATT TGAAAGGATT WMATTATAAA GAACAAAGTC
1681 TCCTAATCTT TGATATAGCA TTGACATACT TTTTAAATAT ACAAGGCATA GAATATGGCC
1741 ATTTCTGTTA AATCATATAT TCCCAACTGG TTATTAATCT AACAATTCAG AATTTTGAGT
1801 AATTGCTTTT GCATCAGATT ATTTACTTCA GTGCTCTCAA TTATGATGGT GCATTAGAAC
1861 CATCTGGGTT AACATTTGTT TTTTATTACC AATACCTAGG CTCCAACCAA GTACAGTGAA
1921 ACTGGAATGT ACAGAGTGGA CAATGGAACG AAGGAGAACA AGACCAAAGG ACATTTTATT
1981 TTTATCTGTA TCAGTGGGTC AAAGTCCTTT CAGAAGGAGC ATATAGTGGA CCTAGGTGAT
2041 TGGTCAATTT ATCCATCAAA GAGGCACACA CCGAATTAGC ATGGAGTGTT ATAAAAGGCT
2101 TGGAGTGCAA GCTCATGGTT GTCTTAACAA GAAGAGAAGG CTTCAATGGA TTCTCTTGTG
2161 GTCCTTGTGC TCTGTCTCTC ATGTTTGCTT CTCCTTTCAC TCTGGAGACA GAGCTCTGGG
2221 AGAGGAAAAC TCCCTCCTGG CCCCACTCCT CTCCCAGTGA TTGGAAATAT CCTACAGATA
2281 GGTATTAAGG ACATCAGCAA ATCCTTAACC AATGTAAGTA TGCTCCTTCA GTGGCTTGCA
2341 AAAGGTAAGT AAATTCACCT GTATTTTTTA AATAAAGTGT ATCCCTAGAG GTACATGTTA
2401 CAAGAGGTAA TGGTAAAGTA AAATACTTTG AAAGGCTT
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DETECTION OF CYP3A4 AND CYP2C9 POLYMORPHISMS

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[0001] The present invention is directed to methods of preparing biological samples for nucleic acid analysis using oligonucleotide primers suitable for amplification of the genes encoding the drug-metabolizing cytochrome P450 enzymes CYP3A4 and CYP2C19.

BACKGROUND OF THE INVENTION

[0002] Xenobiotics are pharmacologically, endocrinologically, or toxicologically active substances foreign to a biological system. Most xenobiotics, including pharmaceutical agents, are metabolized through two successive reactions. Phase I reactions (functionalization reactions), include oxidation, reduction, and hydrolysis, in which a derivatizable group is added to the original molecule. Functionalization prepares the drug for further metabolism in phase II reactions. During phase II reactions (conjugative reactions, which include glucoronidation, sulfation, methylation and acetylation), the functionalized drug is conjugated with a hydrophilic group. The resulting hydrophilic compounds are inactive and excreted in bile or urine. Thus, metabolism can result in detoxification and excretion of the active substance. Alternatively, an inert xenobiotic may be metabolized to an active compound. For example, a pro-drug may be converted to a biologically active therapeutic or toxin.

[0003] The cytochrome P450 (CYP) enzymes are involved in the metabolism of many different xenobiotics. CYPs are a superfamily of heme-containing enzymes, found in eukaryotes (both plants and animals) and prokaryotes, and are responsible for Phase I reactions in the metabolic process. In total, over 500 genes belonging to the CYP superfamily have been described and divided into subfamilies, CYP1-CYP27. In humans, more than 35 genes and 7 pseudogenes have been identified. Members of three CYP gene families, CYP1, CYP2, and CYP3, are responsible for the majority of drug metabolism. The human CYPs which are of greatest clinical relevance for the metabolism of drugs and other xenobiotics are CYP1A2, CYP2A6, CYP2C9, CYP2C19, CYP2D6, CYP2E1 and CYP3A4. The liver is the major site of activity of these enzymes, however CYPs are also expressed in other tissues.

[0004] The most important drug-metabolizing CYP enzyme is CYP3A4, which is the major CYP expressed in liver. Expression of the gene encoding CYP3A4 (CYP3A4) is inducible by many commonly used drugs, such as dexamethasone, rifampicin, and clotrimazole. CYP3A4 is estimated to metabolize more than 60% of all drugs in clinical use, including calcium channel blockers such as nifedipine, immunosuppressants such as cyclosporin A, macrolide antibiotics such as erythromycin, and steroid hormones. In addition, CYP3A4 metabolizes some carcinogens, and may be implicated in an individual's susceptibility to such toxins.

[0005] The existence of more than one form of the CYP3A4 enzyme is caused by polymorphisms in the gene which encodes the CYP3A4 enzyme (the gene being denoted in italics, as CYP3A4). In fact, almost 20 polymorphisms in the CYP3A4 gene have been described (see http://www.imm.ki.se/cypalleles/ for listing). The distribution of particular CYP3A4 polymorphisms differs among ethnic groups, however, concomitant differences in CYP3A4 activity and responses to drugs which are CYP3A4 substrates remain to be investigated. CYP3A4*1A is the wild type gene, corresponding to the cDNA having GenBank Accession No. A18907 and the genomic DNA having GenBank Accession No. AF280107. A number of mutations in the 5' untranslated region of CYP3A4 have been described. CYP3A4*1B is an A to G substitution at position −392. CYP3A4*1C is a T to G substitution at position −444. CYP3A4*1D is a C to A substitution at position −62. CYP3A4*1E is a T to A substitution at position −369. CYP3A4*1F is a C to G substitution at −747. The 5' flanking region of CYP3A4 is set forth in SEQ ID NO: 1 and in **FIG. 1**.

[0006] WO 01/20025 discloses single nucleotide polymorphisms in various exons, introns, and in the 3' UTR of CYP3A4, as well as oligonucleotides for use in diagnosing and treating abnormal expression and/or function of this gene. WO 00/24926 discloses oligonucleotides for use in detecting an A to G point mutation at position −290 of CYP3A4. WO 99/13106 discloses polymorphisms in CYP3A4, including an A to G substitution at position −392 of the promoter, at the 7th position of the 10 bp NFSE, within oligonucleotides having sequences ACAAGGGCAAGAGAGAGGC (SEQ ID NO:2) and ACAAGGGCAGGAGAGAGGC (SEQ ID NO:3), with polymorphic variants indicated in bold type.

[0007] U.S. Pat. No. 6,174,684 and corresponding WO 00/09752 disclose an A to G variant in the nifedipine-specific regulatory element located at positions −287 to −296 of CYP3A4, which is associated with increased risk of prostate cancer and with increased risk of developing leukemia after administration of an epipodophyllotoxin. U.S. Pat. No. 6,174,684 also discloses the oligonucleotides AGGGCAAGAG (SEQ ID NO:4) and AGGGCAGGAG (SEQ ID NO:5), with polymorphic variants indicated in bold type. Rebbeck, et al. (1998) *J. Natl. Cancer Inst.* 90, 1225-1229 also describes this association between prostate cancer, leukemia, and the A to G mutation.

[0008] Kuehl, et al. (2001) *Nature Genetics* 27, 383-391 discloses mutations at positions −341, −288, and −43 of the CYP3A4 promoter, none of which were associated with altered CYP3A4 activity. Kuehl, et al. also discloses differential distribution of these polymorphisms among Caucasians and African Americans.

[0009] A second important CYP enzyme is CYP2C9, which is active in hydroxylation of such drugs as tolbutamide, phenytoin, S-warfarin, diclofenac, ibuprofen, and losarten. The sequence of CYP2C9 is set forth in SEQ ID NO:6. Six variants in CYP2C9 are described on the CYP web site, and another six variant designations are listed without descriptions. The CYP2C9*1 variant is designated as the wild type. Four of the five polymorphic CYP2C9 forms described contain mutations in the coding regions of the gene that result in decreased in vitro activity, and the remaining variant, CYP2C9*6, is a deletion of an A at position 818 which results in a frame shift.

[0010] WO00/12757 discloses primer extension assays and kits for detection of the single nucleotide polymorphisms CYP2C9*2 and CYP2C9*3, both of which result in amino acid substitutions.

[0011] On the basis of ability of metabolize a marker drug such as nifedipine for CYP3A4 or S-warfarin for CYP2C9, individuals may be characterized as poor metabolizers (PM), intermediate metabolizers (IM), extensive metabolizers (EM) or ultra extensive metabolizers (UEM or UM) for CYP3A4 or CYP2C9 substrates, respectively. Poor metabolizers retain the substrate in their bodies for a relatively long period of time, and are susceptible to toxicity and side effects at "normal" dosages. Ultraextensive metabolizers clear the substrate from their bodies quickly, and require higher than "normal" dosages to achieve a therapeutic effect. Intermediate and extensive metabolizers retain the substrate in their bodies for times between those of PMs and UEMs, and are more likely to respond to "normal" dosages of the drug. However, individuals characterized as IM or EM may differ in drug clearance by as much as 10-fold, and variations in toxicity, side effects, and efficacy for a particular drug may occur among these individuals. However, administration of such drugs to determine an individual's metabolic capacity may in itself be dangerous, exposing the individual to potential toxic side effects.

[0012] A need remains for methods of preparing biological samples that contain the 5' flanking regions of CYP3A4 or CYP2C9, so that this information may be used to predict differential capacities for metabolizing CYP3A4 and CYP2C9 substrates among individuals.

SUMMARY OF THE INVENTION

[0013] The present inventors have discovered a novel single nucleotide polymorphism in the 5' flanking region of CYP3A4, and six novel polymorphisms in the 5' flanking region of CYP2C9. Oligonucleotides have been devised for amplification of the polymorphic regions corresponding to these polymorphisms. These oligonucleotides may be used to prepare biological samples for further analysis of the 5' flanking regions of these genes. The inventors have also devised sequence determination oligonucleotides for use as probes for the novel single nucleotide polymorphisms in CYP3A4 and CYP2C9.

[0014] In one embodiment, the invention provides an oligonucleotide primer pair suitable for amplifying a polymorphic region of a 5' flanking region of a CYP3A4 gene, wherein the polymorphic region corresponds to position 461 of SEQ ID NO:1, which position may also be described as position −644 from the transcription start site of the CYP3A4 gene.

[0015] In another embodiment, the invention provides a sequence determination oligonucleotide for detecting a polymorphic site in a 5' flanking region of a CYP3A4 gene, said oligonucleotide being complementary to the polymorphic region corresponding to position 461 of SEQ ID NO:1.

[0016] In another embodiment, the invention provides a kit for amplification and/or detection of a polymorphic region of the 5' flanking region of a CYP3A4 gene, said kit comprising at least one oligonucleotide primer pair capable of amplifying the region corresponding to position 461 of SEQ ID NO:1.

[0017] In another embodiment, the invention provides an oligonucleotide primer pair suitable for amplifying a polymorphic region of a 5' flanking region of a CYP2C9 gene, wherein the polymorphic region corresponds to position 957 of SEQ ID NO:6; position 1049 of SEQ ID NO:6; position 1164 of SEQ ID NO:6; position 1526 of SEQ ID NO:6; position 1661 of SEQ ID NO:6; and position 1662 of SEQ ID NO:6. Position 957 of SEQ ID NO:6 may also be described as position −1189 from the transcription start site of the CYP3C9 gene; position 1049 of SEQ ID NO:6 may also be described as position −1097 from the transcription start site; position 1164 of SEQ ID NO:6 may also be described as position −982 from the transcription start site; position 1526 of SEQ ID NO:6 may also be described as position −620 from the transcription start site; position 1661 of SEQ ID NO:6 may also be described as position −485 from the transcription start site; and position 1662 of SEQ ID NO:6 may also be described as position −484 from the transcription start site.

[0018] In yet another embodiment, the invention provides a sequence determination oligonucleotide for detecting a polymorphic site in a 5' flanking region of a CYP2C9 gene, said oligonucleotide comprising a sequence selected from the group consisting of an oligonucleotide complementary to the polymorphic region corresponding to position 957 of SEQ ID NO:6; an oligonucleotide complementary to the polymorphic region corresponding to position 1049 of SEQ ID NO:6; an oligonucleotide complementary to the polymorphic region corresponding to position 1164 of SEQ ID NO:6; an oligonucleotide complementary to the polymorphic region corresponding to position 1526 of SEQ ID NO:6; an oligonucleotide complementary to the polymorphic region corresponding to position 1661 of SEQ ID NO:6; and an oligonucleotide complementary to the polymorphic region corresponding to position 1662 of SEQ ID NO:6.

[0019] In another embodiment, the invention provides a kit for amplification and/or detection of a polymorphic region corresponding to at least one polymorphic region in the 5' flanking region of the CYP2C9 gene, said region being selected from the group consisting of position 957 of SEQ ID NO:6; position 1049 of SEQ ID NO:6; position 1164 of SEQ ID NO:6; position 1526 of SEQ ID NO:6; position 1661 of SEQ ID NO:6; and position 1662 of SEQ ID NO:6.

BRIEF DESCRIPTION OF THE DRAWINGS

[0020] **FIG. 1** shows the sequence of the 5' flanking region of the CYP3A4 gene as set forth in SEQ ID NO: 1, with the novel polymorphic site underlined and highlighted in bold.

[0021] **FIG. 2** shows the sequence of the 5' flanking region of the CYP2C9 gene as set forth in SEQ ID NO:6, with the novel polymorphic sites underlined and highlighted in bold.

DETAILED DESCRIPTION OF THE INVENTION

[0022] The U.S. patents and publications referenced herein are hereby incorporated by reference.

[0023] For the purposes of the invention, certain terms are defined as follows. "Gene" is defined as the genomic sequence of the CYP2C19 gene. "Oligonucleotide" means a nucleic acid molecule preferably comprising from about 8 to about 50 covalently linked nucleotides. More preferably, an oligonucleotide of the invention comprises from about 8 to about 35 nucleotides. Most preferably, an oligonucleotide of the invention comprises from about 10 to about 25 nucle-

otides. In accordance with the invention, the nucleotides within an oligonucleotide may be analogs or derivatives of naturally occurring nucleotides, so long as oligonucleotides containing such analogs or derivatives retain the ability to hybridize specifically within the polymorphic region containing the targeted polymorphism. Analogs and derivatives of naturally occurring oligonucleotides within the scope of the present invention are exemplified in U.S. Pat. Nos. 4,469,863; 5,536,821; 5,541,306; 5,637,683; 5,637,684; 5,700,922; 5,717,083; 5,719,262; 5,739,308; 5,773,601; 5,886,165; 5,929,226; 5,977,296; 6,140,482; WO 00/56746; WO 01/14398, and the like. Methods for synthesizing oligonucleotides comprising such analogs or derivatives are disclosed, for example, in the patent publications cited above and in U.S. Pat. Nos. 5,614,622; 5,739,314; 5,955,599; 5,962,674; 6,117,992; in WO 00/75372, and the like. The term "oligonucleotides" as defined herein also includes compounds which comprise the specific oligonucleotides disclosed herein, covalently linked to a second moiety. The second moiety may be an additional nucleotide sequence, for example, a tail sequence such as a polyadenosine tail or an adaptor sequence, for example, the phage M13 universal tail sequence, and the like. Alternatively, the second moiety may be a non-nucleotidic moiety, for example, a moiety which facilitates linkage to a solid support or a label to facilitate detection of the oligonucleotide. Such labels include, without limitation, a radioactive label, a fluorescent label, a chemiluminescent label, a paramagnetic label, and the like. The second moiety may be attached to any position of the specific oligonucleotide, so long as the oligonucleotide retains its ability to hybridize to the polymorphic regions described herein.

[0024] A polymorphic region as defined herein is a portion of a genetic locus that is characterized by at least one polymorphic site. A genetic locus is a location on a chromosome which is associated with a gene, a physical feature, or a phenotypic trait. A polymorphic site is a position within a genetic locus at which at least two alternative sequences have been observed in a population. A polymorphic region as defined herein is said to "correspond to" a polymorphic site, that is, the region may be adjacent to the polymorphic site on the 5' side of the site or on the 3' side of the site, or alternatively may contain the polymorphic site. A polymorphic region includes both the sense and antisense strands of the nucleic acid comprising the polymorphic site, and may have a length of from about 100 to about 5000 base pairs. For example, a polymorphic region may be all or a portion of a regulatory region such as a promoter, 5' UTR, 3' UTR, an intron, an exon, or the like. A polymorphic or allelic variant is a genomic DNA, cDNA, mRNA or polypeptide having a nucleotide or amino acid sequence that comprises a polymorphism. A polymorphism is a sequence variation observed at a polymorphic site, including nucleotide substitutions (single nucleotide polymorphisms or SNPs), insertions, deletions, and microsatellites. Polymorphisms may or may not result in detectable differences in gene expression, protein structure, or protein function. Preferably, a polymorphic region of the present invention has a length of about 1000 base pairs. More preferably, a polymorphic region of the invention has a length of about 500 base pairs. Most preferably, a polymorphic region of the invention has a length of about 200 base pairs.

[0025] A haplotype as defined herein is a representation of the combination of polymorphic variants in a defined region within a genetic locus on one of the chromosomes in a chromosome pair. A genotype as used herein is a representation of the polymorphic variants present at a polymorphic site.

[0026] The PCR primer pairs of the invention are capable of amplifying the polymorphic region corresponding to position 461 of SEQ ID NO: 1, or any of the polymorphic regions corresponding to position 957 of SEQ ID NO:6; position 1049 of SEQ ID NO:6; position 1164 of SEQ ID NO:6; position 1526 of SEQ ID NO:6; position 1661 of SEQ ID NO:6; and position 1662 of SEQ ID NO:6. Specific oligonucleotide primer pairs of the invention, for amplifying position 461 of SEQ ID NO:1, may comprise sequences selected from the group consisting of SEQ ID NO:7 and SEQ ID NO:8; and SEQ ID NO;9 and SEQ ID NO: 10. For amplifying only position 957 of SEQ ID NO:6, an oligonucleotide primer pair comprising the sequences set forth in SEQ ID NO: 19 and SEQ ID NO:20 may be used. Alternatively, positions 957 and 1049 of SEQ ID NO:6 may be amplified using an oligonucleotide primer pair comprising the sequences set forth in SEQ ID NO:21 and SEQ ID NO:22; or positions 957,1049, and 1164 may be amplified using an oligonucleotide primer pair comprising the sequences set forth in SEQ ID NO:23 and SEQ ID NO:24. Position 1164 of SEQ ID NO:6 may also be amplified using an oligonucleotide primer pair comprising the sequences set forth in SEQ ID NO:25 and SEQ ID NO:26. Positions 1526, 1661, and 1662 of SEQ ID NO:6 may be amplified using an oligonucleotide primer pair comprising the sequences set forth in SEQ ID NO:27 and SEQ ID NO:28. Positions 1661 and 1662 of SEQ ID NO:6 may be amplified using an oligonucleotide primer pair selected from the group consisting of an oligonucleotide primer pair comprising the sequences set forth in SEQ ID NO:29 and SEQ ID NO:30 and an oligonucleotide primer pair comprising the sequences set forth in SEQ ID NO:31 and SEQ ID NO:32.

[0027] Each of the PCR primer pairs of the invention may be used in any PCR method. For example, a PCR primer pair of the invention may be used in the methods disclosed in U.S. Pat. Nos. 4,683,195; 4,683,202, 4,965,188; 5,656,493; 5,998,143; 6,140,054; WO 01/27327; WO 01/27329; and the like. The PCR pairs of the invention may also be used in any of the commercially available machines that perform PCR, such as any of the GeneAmp® Systems available from Applied Biosystems.

[0028] The oligonucleotides of the invention may be used to determine the sequence of the polymorphic regions of SEQ ID NO: 1 or SEQ ID NO:6 as defined herein. In one embodiment, an oligonucleotide of the invention comprises a sequence selected from the group consisting of SEQ ID NO:11; SEQ ID NO:12; SEQ ID NO:13; SEQ ID NO:14; SEQ ID NO:15; SEQ ID NO: 16; SEQ ID NO:17; and SEQ ID NO:18, for determining the sequence of the novel polymorphic region of CYP3A4 corresponding to position 461 of SEQ ID NO:1. In another embodiment, for determining the sequence of the polymorphic region of CYP2C9 corresponding to position 957 of SEQ ID NO:6, an oligonucleotide of the invention comprises a sequence selected from the group consisting of SEQ ID NO:33; SEQ ID NO:34; SEQ ID NO:43; SEQ ID NO:44; SEQ ID NO:53; SEQ ID NO:58; SEQ ID NO:63; and SEQ ID NO:68. In another embodiment, for determining the sequence of the polymorphic region of CYP2C9 corresponding to position 1049 of

SEQ ID NO:6, an oligonucleotide of the invention comprises a sequence selected from the group consisting of SEQ ID NO:35; SEQ ID NO:36; SEQ ID NO:45; SEQ ID NO:46; SEQ ID NO:54; SEQ ID NO:59; SEQ ID NO:64; and SEQ ID NO:69. In another embodiment, for determining the sequence of the polymorphic region of CYP2C9 corresponding to position 1164 of SEQ ID NO:6, an oligonucleotide of the invention comprises a sequence selected from the group consisting of SEQ ID NO:37; SEQ ID NO:38; SEQ ID NO:45; SEQ ID NO:48; SEQ ID NO:55; SEQ ID) NO:60; SEQ ID) NO:65; and SEQ ID NO:70. In another embodiment, for determining the sequence of the polymorphic region of CYP2C9 corresponding to position 1526 of SEQ ID NO:6, an oligonucleotide of the invention comprises a sequence selected from the group consisting of SEQ ID NO:39; SEQ ID NO:40; SEQ ID NO:49; SEQ ID NO:50; SEQ if) NO:56; SEQ ID NO:61; SEQ ID NO:66; and SEQ ID NO:71. In another embodiment, for determining the sequences of the polymorphic region of CYP2C9 corresponding to either of positions 1661 or 1662 of SEQ ID NO:6, an oligonucleotide of the invention comprises a sequence selected from the group consisting of SEQ ID NO:41; SEQ ID NO:42; SEQ ID NO:51; SEQ ID NO:52; SEQ ID NO:57; SEQ ID NO:62; SEQ ID NO:67; and SEQ ID NO:72.

[0029] Those of ordinary skill will recognize that oligonucleotides complementary to the polymorphic regions described herein must be capable of hybridizing to the polymorphic regions under conditions of stringency such as those employed in primer extension-based sequence determination methods, restriction site analysis, nucleic acid amplification methods, ligase-based sequencing methods, methods based on enzymatic detection of mismatches, microarray-based sequence determination methods, and the like. The oligonucleotides of the invention may be synthesized using known methods and machines, such as the ABI™3900 High Throughput DNA Synthesizer and the Expedite™8909 Nucleic Acid Synthesizer, both of which are available from Applied Biosystems (Foster City, Calif.).

[0030] The oligonucleotides of the invention may be used, without limitation, as in situ hybridization probes or as components of diagnostic assays. Numerous oligonucleotide-based diagnostic assays are known. For example, primer extension-based nucleic acid sequence detection methods are disclosed in U.S. Pat. Nos. 4,656,127; 4,851,331; 5,679,524; 5,834,189; 5,876,934; 5,908,755; 5,912,118; 5,976,802; 5,981,186; 6,004,744; 6,013,431; 6,017,702; 6,046,005; 6,087,095; 6,210,891; WO 01/20039; and the like. Primer extension-based nucleic acid sequence detection methods using mass spectrometry are described in U.S. Pat. Nos. 5,547,835; 5,605,798; 5,691,141; 5,849,542; 5,869,242; 5,928,906; 6,043,031; 6,194,144, and the like. The oligonucleotides of the invention are also suitable for use in ligase-based sequence determination methods such as those disclosed in U.S. Pat. Nos. 5,679,524 and 5,952,174, WO 01/27326, and the like. The oligonucleotides of the invention may be used as probes in sequence determination methods based on mismatches, such as the methods described in U.S. Pat. Nos. 5,851,770; 5,958,692; 6,110,684; 6,183,958; and the like. In addition, the oligonucleotides of the invention may be used in hybridization-based diagnostic assays such as those described in U.S. Pat. Nos. 5,891,625; 6,013,499; and the like.

[0031] The oligonucleotides of the invention may also be used as components of a diagnostic microarray. Methods of making and using oligonucleotide microarrays suitable for diagnostic use are disclosed in U.S. Pat. Nos. 5,492,806; 5,525,464; 5,589,330; 5,695,940; 5,849,483; 6,018,041; 6,045,996; 6,136,541; 6,142,681; 6,156,501; 6,197,506; 6,223,127; 6,225,625; 6,229,911; 6,239,273; WO 00/52625; WO 01/25485; WO 01/29259; and the like.

[0032] The invention is also embodied in a kit comprising at least one oligonucleotide primer pair of the invention. When the kit is used for amplification and detection of CYP3A4 polymorphisms, it will comprise an oligonucleotide primer pair suitable for amplification of the polymorphic region corresponding to position 461 of SEQ ID NO:1.

[0033] Specific primer pairs in this embodiment are selected from the group consisting of SEQ ID NO:7 and SEQ ID NO:8; and SEQ ID NO;9 and SEQ ID NO:10. This embodiment of the kit of the invention may optionally comprise a sequence determination oligonucleotide selected from the group consisting of SEQ ID NO: 11; SEQ ID NO: 12; SEQ ID NO:13; SEQ ID NO: 14; SEQ ID NO:15; SEQ ID NO:16; SEQ ID NO:17; and SEQ ID NO:18.

[0034] When the kit of the invention is used for amplification and detection of polymorphisms in the 5' flanking region of CYP2C9, it will comprise at least one oligonucleotide primer pair, wherein the primer pair is capable of amplifying a polymorphic region selected from the group consisting of the polymorphic region corresponding to position 957 of SEQ ID NO:6; the polymorphic region corresponding to position 1049 of SEQ ID NO:6; the polymorphic region corresponding to position 1164 of SEQ ID NO:6; the polymorphic region corresponding to position 1526 of SEQ ID NO:6; the polymorphic region corresponding to position 1661 of SEQ ID NO:6; and the polymorphic region corresponding to position 1662 of SEQ ID NO:6. This embodiment may optionally further comprise a sequence determination oligonucleotide for detecting a polymorphic variant at any or all of the polymorphic sites corresponding to positions 957, 1049, 1164, 1526, 1661 and 1662 of SEQ ID NO:6.

[0035] The kit of the invention may also comprise a polymerizing agent, for example, a thermostable nucleic acid polymerase such as those disclosed in U.S. Pat. Nos. 4,889,818; 6,077,664, and the like. The kit of the invention may also comprise chain elongating nucleotides, such as dATP, dTTP, dGTP, dCTP, and dITP, including analogs of dATP, dTTP, dGTP, dCTP and dITP, so long as such analogs are substrates for a thermostable nucleic acid polymerase and can be incorporated into a growing nucleic acid chain. The kit of the invention may also include chain terminating nucleotides such as ddATP, ddTTP, ddGTP, ddCTP, and the like. In a preferred embodiment, the kit of the invention comprises at least two oligonucleotide primer pairs, a polymerizing agent, chain elongating nucleotides, at least two sequence determination oligonucleotides and at least one chain terminating nucleotide. The kit of the invention may optionally include buffers, vials, microtiter plates, and instructions for use.

[0036] The examples set forth below are provided as illustration and are not intended to limit the scope and spirit of the invention as specifically embodied therein.

EXAMPLE 1

IDENTIFICATION OF CYP3A4 POLYMORPHISM

[0037] The study was performed in accordance with the principles stated in the Declaration of Helsinki as reviewed in Tokyo 1975 and Venice 1983, Hong Kong 1989 and Somerset West 1996. Ten samples (Swedish Caucasians) were selected and used for identification of polymorphisms in the 5' flanking region of CYP3A4.

[0038] White blood cells isolated from a blood sample drawn from the brachial vein serve as the source of the genomic DNA for the analyses. The DNA was extracted by guanidine thiocyanate method or Q1Aamp Blood Kit (QIAGEN, Venlo, The Netherlands). The genes included in the study were amplified by PCR and the DNA sequences were determined by full sequencing. All genetic analyses were performed according to Good Laboratory Practice and Standard Operating Procedures. Case Report Forms were designed and used for clinical and genetic data collection. Data was entered and stored in a relational database at Gemini Genomics AB, Uppsala. To secure consistency between the Case Report Forms and the database, data was checked either by double data entry or proofreading. After a Clean File was declared the database was protected against changes. By using the program Stat/Transfer™ the database was transferred to SAS data sets. The SAS™ system was used for tabulations and statistical evaluations. Genotypes were also correlated against the metabolic ratio.

[0039] PCR-fragments were amplified with TaqGOLD polymerase (Applied Biosystems) using Robocycler (Stratagene) or GeneAmp PCR system 9700 (Applied Biosystems). Preferentially, the amplified fragments were 300-400 bp, and the region to be read did not exceed 300 bp. PCR reactions were carried out according to the basic protocol set forth in Table 1, with modifications as indicated in Table 2 for specific primer pairs, which are shown in Table 3. For the GeneAmp PCR 9700 machine the profile used was 10 minutes at 95°, 40×(45 seconds at 90°, 45 seconds at 60°, 45 seconds at 72°), 5 minutes at 72° and 22° until removed.

TABLE 1

Solution	Stock Concentration	PCR (μl)
H_2O		33.2
PCR buffer	10x	5.0
$MgCl_2$	25 mM	2.0
dNTP	2.5 mM	2.5
primer 1	10 μM	1.0
primer 2	10 μM	1.0
Taq-gold polymerase	5 μ/μl	0.3
DNA samples	2 ng/μl	5.0
TOTAL		50.0

[0040]

TABLE 2

SEQ ID NO:s	Polymorphic Site	Modification from basic protocol (Table 1)	Detection method
7, 8	461	62° annealing temperature	Full sequencing
9, 10	461	3 μl $MgCl_2$, 58° annealing temperature, 50 cycles	Full sequencing

[0041]

TABLE 3

Polymorphic Site		Primer Pair
461	SEQ ID NO:7	CCAGCCTGAAAGTGCAGAGA
	SEQ ID NO:8	TCTTAGAGTCTTTCCTCACCAAACT
461	SEQ ID NO:9	CATGCCCTGTCTCTCCTTTA
	SEQ ID NO:10	CCATCCCCTTCATGCAATC

[0042] The optimized condition specified in Table 2 were required to distinguish CYP3A4 from the closely related gene-family members CYP3A5, and CYP3A7. Use of the basic protocol will lead to problems when amplifying CYP3A4-specific amplicons of 300-400 bp containing the polymorphisms of interest, unless a nested PCR approach is carried out. The nested PCR approach was not used because of the high risk of contamination when using a nested PCR approach and the high risk of typing errors as a consequence. The modifications shown in Table 2 were optimized and reaction parameters were balanced in such a way that nested PCR was avoided.

[0043] For full sequencing, one of the PCR-primers in a primer pair was designed for sequencing by addition of a 29 nucleotide tail complementary to M13 at its 5'-end, namely the nucleotides AGTCACGACGTTGTAAAACGACGGCCAGT. Thus, the entire PCR-product was sequenced from the tailed PCR-primer.

[0044] The additional oligonucleotides set forth in Tables 4 through 7 were identified as being suitable for detection of the SNP at positions 461 of the 5' flanking region of the CYP3A4 gene as depicted in SEQ ID NO: 1.

[0045] Table 4 sets forth oligonucleotides representing the coding (sense) strand complementary to the polymorphic region corresponding to the novel polymorphism found in the study population. The underlined letter indicates polymorphic position in the sequence context. All sequences are shown in 5' to 3' direction.

TABLE 4

Polymorphic Site		Sequence	Note
461	SEQ ID NO:11:	AGCAC<u>C</u>CTGGT	C variant
	SEQ ID NO:12:	AGCAC<u>G</u>CTGGT	G variant

[0046] Table 5 sets forth oligonucleotides representing the non-coding (anti-sense) strand complementary to the polymorphic region corresponding to the novel polymorphism found in the study population. The underlined letter indicates polymorphic position in the sequence context. All sequences are shown in 5' to 3' direction.

TABLE 5

Polymorphic Site	Sequence	Note
461	SEQ ID NO:13: ACCAGGGTGCT	Antisense G variant
	SEQ ID NO:14: ACCAGCGTGCT	Antisense C variant

[0047] The sequences of Table 6 represent the 5'-sequence to the novel polymorphic site on the coding (sense) strand (SEQ ID NO: 15) and non-coding (anti-sense) strand (SEQ ID NO:s 16). All sequences are shown in 5' to 3' direction.

TABLE 6

Polymorphic Site	Sequence	Note
461	SEQ ID NO:15: GTGTGTACAGC	Sense 5'
	SEQ ID NO:16: GCTGTACACAC	Antisense 5'

[0048] The sequences of Table 7 represent the 3'-sequence to the novel polymorphic site on the non-coding (anti-sense) strand (SEQ ID NO: 17) and the coding (sense) strand (SEQ ID NO:18). All sequences are shown in 5' to 3' direction.

TABLE 9

Polymorphic Site	Sequence	Note
461	SEQ ID NO:17: TGGTCCCTACC	Antisense 3'
	SEQ ID NO:18: GGTAGGGACCA	Sense 3'

EXAMPLE 2

IDENTIFICATION OF CYP2C9 POLYMORPHISMS

[0049] The study was performed in accordance with the principles stated in the Declaration of Helsinki as reviewed in Tokyo 1975 and Venice 1983, Hong Kong 1989 and Somerset West 1996. Ten samples (Swedish Caucasians) were selected and used for identification of polymorphisms in the 5' flanking region of CYP2C9.

[0050] White blood cells isolated from a blood sample drawn from the brachial vein serve as the source of the genomic DNA for the analyses. The DNA is extracted by guanidine thiocyanate method or Q1Aamp Blood Kit (QIAGEN, Venlo, The Netherlands). The genes included in the study were amplified by PCR and the DNA sequences were determined by full sequencing. All genetic analyses were performed according to Good Laboratory Practice and Standard Operating Procedures. Case Report Forms were designed and used for clinical and genetic data collection. Data was entered and stored in a relational database at Gemini Genomics AB, Uppsala. To secure consistency between the Case Report Forms and the database, data was checked either by double data entry or proofreading. After a Clean File was declared the database was protected against changes. By using the program Stat/Transfer™ the database was transferred to SAS data sets. The SAS™ system was used for tabulations and statistical evaluations. Genotypes were also correlated against the metabolic ratio.

[0051] PCR-fragments were amplified with TaqGOLD polymerase (Applied Biosystems) using Robocycler (Stratagene) or GeneAmp PCR system 9700 (Applied Biosystems). Preferentially, the amplified fragments were 300-400 bp, and the region to be read did not exceed 300 bp. PCR reactions were carried out according to the basic protocol set forth in Table 10, with modifications as indicated in Table 11 for specific primer pairs, which are shown in Table 12. For the GeneAmp PCR 9700 machine the profile used was 10 minutes at 95°, 40×(45 seconds at 90°, 45 seconds at 60°, 45 seconds at 72°), 5 minutes at 72° and 22° until removed.

TABLE 10

Solution	Stock Concentration	PCR (μl)
H_2O		33.2
PCR buffer	10x	5.0
$MgCl_2$	25 mM	2.0
dNTP	2.5 mM	2.5
primer 1	10 μM	1.0
primer 2	10 μM	1.0
Taq-gold polymerase	5 μ/μl	0.3
DNA samples	2 ng/μl	5.0
TOTAL		50.0

[0052]

TABLE 11

SEQ ID NO:s	Polymorphic Site	Modification from basic protocol (Table 10)	Detection method
19, 20	957	58° annealing temperature	Full sequencing
21, 22	957 & 1049	3 μl $MgCl_2$, 62° annealing temperature	Full sequencing
23, 24	957, 1049 & 1164	58° annealing temperature	Full sequencing
25, 26	1164	3 μl $MgCl_2$, 62° annealing temperature, 50 cycles	Full sequencing
27, 28	1526, 1661 & 1662		Full sequencing
29, 30	1661 & 1662	3 μl $MgCl_2$, 62° annealing temperature, 50 cycles	Full sequencing
31, 32	1661 & 1662		Full sequencing

[0053]

TABLE 12

Polymorphic Site	Primer Pair	
957	SEQ ID NO:19	CACTAGGGAATTTAGAACAAATATG
	SEQ ID NO:20	GCACAGAAAGCAAAGGAAATTAT
957 & 1049	SEQ ID NO:21	TGTATTTAGATCCTCAACTCAG-TATGT
	SEQ ID NO:22	GGATCTCCCTTCTCCATCACT
957, 1049 & 1164	SEQ ID NO:23	GGTCCATTTAGTGATTTCCCTAC
	SEQ ID NO:24	ATACACCACATTTATTCTGTTCATA

TABLE 12-continued

Polymorphic Site	Primer Pair	
1164	SEQ ID NO:25	CCAAATTTTTCCCTCAGTTACA
	SEQ ID NO:26	TTGGTGCCACACAGCTCATA
1526, 1661 & 1662	SEQ ID NO:27	GCCTTCAGGAATTTTTTTTA
	SEQ ID NO:28	CCAGTTGGGAATATATGATTTAACA
1661 & 1662	SEQ ID NO:29	GCTGCTGTATTTTTAGTAGGCTATA
	SEQ ID NO:30	CGTTCCATTGTCCACTCTGTAC
1661 & 1662	SEQ ID NO:31	TCAAGGCAGCTCTGGTGTAA
	SEQ ID NO:32	AGTTGGGAATATATGATTTAACAGA

[0054] The optimized condition specified in Table 11 were required to distinguish CYP2C9 from the closely related gene-family members CYP2C8, CYP2C18 and CYP2C19. Use of the basic protocol will lead to problems when amplifying CYP2C9-specific amplicons of 300-400 bp containing the polymorphisms of interest, unless a nested PCR approach is carried out. The nested PCR approach was not used because of the high risk of contamination when using a nested PCR approach and the high risk of typing errors as a consequence. The modifications shown in Table 11 were optimized and reaction parameters were balanced in such a way that nested PCR was avoided.

[0055] For full sequencing, one of the PCR-primers in a primer pair was designed for sequencing by addition of a 29 nucleotide tail complementary to M13 at its 5'-end, namely the nucleotides AGTCACGACGTTGTAAAACGACGGCCAGT. Thus, the entire PCR-product was sequenced from the tailed PCR-primer. The additional oligonucleotides set forth in Tables 13 through 16 were identified as being suitable for detection of the SNPs at positions 957, 1049, 1164, 1526, 1661 and/or 1662 of the 5' flanking region of the CYP2C9 gene as depicted in SEQ ID NO:6.

[0056] Table 13 sets forth oligonucleotides representing the coding (sense) strand complementary to the polymorphic region corresponding to the polymorphisms found in the study population. The underlined letter indicates polymorphic position in the sequence context. All sequences are shown in 5' to 3' direction.

TABLE 13

Polymorphic Site	Sequence		Note
957	SEQ ID NO:33:	ATCTT<u>C</u>TATTG	C variant
	SEQ ID NO:34:	ATCTT<u>T</u>TATTG	T Variant
1049	SEQ ID NO:35:	ACAAT<u>A</u>GAAAG	A variant
	SEQ ID NO:36:	ACAAT<u>G</u>GAAAG	G variant
1164	SEQ ID NO:37:	ATGGA<u>G</u>AAGGG	G variant
	SEQ ID NO:38:	ATGGA<u>A</u>AAGGG	A variant
1526	SEQ ID NO:39:	TTAAT<u>G</u>GTAAA	G variant
	SEQ ID NO:40:	TTAAT<u>T</u>GTAAA	T variant
1661 & 1662	SEQ ID NO:41:	GGATT<u>TC</u>ATTAT	TC variants
	SEQ ID NO:42:	GGATT<u>AA</u>ATTAT	AA variants

[0057] Table 14 sets forth oligonucleotides representing the non-coding (anti-sense) strand complementary to the polymorphic region corresponding to the polymorphisms found in the study population. The underlined letter indicates polymorphic position in the sequence context. All sequences are shown in 5' to 3' direction.

TABLE 14

Polymorphic Site	Sequence		Note
957	SEQ ID NO:43:	CAATA<u>G</u>AAGAT	Antisense G variant
	SEQ ID NO:44:	CAATA<u>A</u>AAGAT	Antisense A variant
1049	SEQ ID NO:45:	CTTTC<u>T</u>ATTGT	Antisense T variant
	SEQ ID NO:46:	CTTTC<u>C</u>ATTGT	Antisense C variant
1164	SEQ ID NO:47:	CCCTT<u>C</u>TCCAT	Antisense C variant
	SEQ ID NO:48:	CCCTT<u>T</u>TCCAT	Antisense T variant
1526	SEQ ID NO:49:	TTTAC<u>C</u>ATTAA	Antisense C variant
	SEQ ID NO:50:	TTTAC<u>A</u>ATTAA	Antisense A variant
1661 & 1662	SEQ ID NO:51:	ATAAT<u>GA</u>AATCC	Antisense GA variants
	SEQ ID NO:52:	ATAAT<u>TT</u>AATCC	Antisense TT variant

[0058] The sequences of Table 15 represent the 5'-sequence to the polymorphic sites on the coding (sense) strand (SEQ ID NO:s 53-57) and non-coding (anti-sense) strand (SEQ ID NO:s 58-67). All sequences are shown in 5' to 3' direction.

TABLE 15

Polymorphic Site	Sequence		Note
957	SEQ ID NO:53:	TACCTCCCATC	Sense 5'
	SEQ ID NO:58:	GATGGGAGGTA	Antisense 5'
1049	SEQ ID NO:54:	AACCAAAAACA	Sense 5'
	SEQ ID NO:59:	TGTTTTTGGTT	Antisense 5'
1164	SEQ ID NO:55:	CTGCAGTGATG	Sense 5'
	SEQ ID NO:60:	CATCACTGCAG	Antisense 5'
1526	SEQ ID NO:56:	TAGGGGGTTTA	Sense 5'
	SEQ ID NO:61:	TAAACCCCCTA	Antisense 5'
1661 & 1662	SEQ ID NO:57:	ATTTGAAAGGA	Sense 5'
	SEQ ID NO:62:	TCCTTTCAAAT	Antisense 5'

[0059] The sequences of Table 16 represent the 3'-sequence to the polymorphic sites on the non-coding (anti-sense) strand (SEQ ID NO:s 68-72) and the coding (sense) strand (SEQ ID NO:s 73-77). All sequences are shown in 5' to 3' direction.

TABLE 16

Polymorphic Site	Sequence	Note
957	SEQ ID NO:63: TGTGGATGCAA	Antisense 3'
	SEQ ID NO:68: TTGCATCCACA	Sense 3'
1049	SEQ ID NO:64: CATGGCTGCTT	Antisense 3'
	SEQ ID NO:69: AAGCAGCCATG	Sense 3'
1164	SEQ ID NO:65: AGGGATCTCCC	Antisense 3'
	SEQ ID NO:70: GGGAGATCCCT	Sense 3'

TABLE 16-continued

Polymorphic Site	Sequence	Note
1526	SEQ ID NO:66: TAAACACCTTT	Antisense 3'
	SEQ ID NO:71: AAAGGTGTTTA	Sense 3'
1661 & 1662	SEQ ID NO:67: TGTTCTTTATA	Antisense 3'
	SEQ ID NO:72: TATAAAGAACA	Sense 3'

[0060]

SEQUENCE LISTING

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<212> TYPE: DNA
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<400> SEQUENCE: 2

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<400> SEQUENCE: 3

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gtccttgtgc tctgtctctc atgtttgctt ctcctttcac tctggagaca gagctctggg 2220

agaggaaaac tccctcctgg ccccactcct ctcccagtga ttggaaatat cctacagata 2280

ggtattaagg acatcagcaa atccttaacc aatgtaagta tgctccttca gtggcttgca 2340

aaaggtaagt aaattcacct gtatttttta aataaagtgt atccctagag gtacatgtta 2400

caagaggtaa tggtaaagta aaatactttg aaaggctt 2438

<210> SEQ ID NO 7
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<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer

<400> SEQUENCE: 7

ccagcctgaa agtgcagaga 20

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<210> SEQ ID NO 8
<211> LENGTH: 25
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer

<400> SEQUENCE: 8

tcttagagtc tttcctcacc aaact 25

<210> SEQ ID NO 9
<211> LENGTH: 20
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer

<400> SEQUENCE: 9

catgccctgt ctctccttta 20

<210> SEQ ID NO 10
<211> LENGTH: 19
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer

<400> SEQUENCE: 10

ccatcccctt catgcaatc 19

<210> SEQ ID NO 11
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the coding strand complementary to the polymorphic site 461

<400> SEQUENCE: 11

agcaccctgg t 11

<210> SEQ ID NO 12
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the coding strand complementary to the polymorphic site 461

<400> SEQUENCE: 12

agcacgctgg t 11

<210> SEQ ID NO 13
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the non-coding strand complementary to the polymorphic site 461

<400> SEQUENCE: 13

accagggtgc t 11

<210> SEQ ID NO 14

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<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the non-coding strand complementary to the polymorphic site 461

<400> SEQUENCE: 14

accagcgtgc t 11

<210> SEQ ID NO 15
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide of the novel polymorphic site 461 on the coding strand

<400> SEQUENCE: 15

gtgtgtacag c 11

<210> SEQ ID NO 16
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide of the novel polymorphic site 461 on the non-coding strand

<400> SEQUENCE: 16

gctgtacaca c 11

<210> SEQ ID NO 17
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide of the novel polymorphic site 461 on the non-coding strand

<400> SEQUENCE: 17

tggtcgctac c 11

<210> SEQ ID NO 18
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide of the novel polymorphic site 461 on the coding strand

<400> SEQUENCE: 18

ggtagggacc a 11

<210> SEQ ID NO 19
<211> LENGTH: 25
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer

<400> SEQUENCE: 19

cactagggaa tttagaacaa atatg 25

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<210> SEQ ID NO 20
<211> LENGTH: 23
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer

<400> SEQUENCE: 20

gcacagaaag caaaggaaat tat 23

<210> SEQ ID NO 21
<211> LENGTH: 27
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer

<400> SEQUENCE: 21

tgtatttaga tcctcaactc agtatgt 27

<210> SEQ ID NO 22
<211> LENGTH: 21
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer

<400> SEQUENCE: 22

ggatctccct tctccatcac t 21

<210> SEQ ID NO 23
<211> LENGTH: 23
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer

<400> SEQUENCE: 23

ggtccattta gtgatttccc tac 23

<210> SEQ ID NO 24
<211> LENGTH: 25
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer

<400> SEQUENCE: 24

atacaccaca tttattctgt tcata 25

<210> SEQ ID NO 25
<211> LENGTH: 22
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer

<400> SEQUENCE: 25

ccaaattttt ccctcagtta ca 22

<210> SEQ ID NO 26
<211> LENGTH: 20
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence

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<220> FEATURE:
<223> OTHER INFORMATION: Primer

<400> SEQUENCE: 26

ttggtgccac acagctcata 20

<210> SEQ ID NO 27
<211> LENGTH: 20
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer

<400> SEQUENCE: 27

gccttcagga attttttta 20

<210> SEQ ID NO 28
<211> LENGTH: 25
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer

<400> SEQUENCE: 28

ccagttggga atatatgatt taaca 25

<210> SEQ ID NO 29
<211> LENGTH: 25
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer

<400> SEQUENCE: 29

gctgctgtat ttttagtagg ctata 25

<210> SEQ ID NO 30
<211> LENGTH: 22
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer

<400> SEQUENCE: 30

cgttccattg tccactctgt ac 22

<210> SEQ ID NO 31
<211> LENGTH: 20
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer

<400> SEQUENCE: 31

tcaaggcagc tctggtgtaa 20

<210> SEQ ID NO 32
<211> LENGTH: 25
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer

<400> SEQUENCE: 32

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agttgggaat atatgattta acaga 25

<210> SEQ ID NO 33
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the coding strand

<400> SEQUENCE: 33

atcttctatt g 11

<210> SEQ ID NO 34
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the coding strand

<400> SEQUENCE: 34

atcttttatt g 11

<210> SEQ ID NO 35
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the coding strand

<400> SEQUENCE: 35

acaatagaaa g 11

<210> SEQ ID NO 36
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the coding strand

<400> SEQUENCE: 36

acaatggaaa g 11

<210> SEQ ID NO 37
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the coding strand

<400> SEQUENCE: 37

atggagaagg g 11

<210> SEQ ID NO 38
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the coding strand

<400> SEQUENCE: 38

atggaaaagg g 11

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<210> SEQ ID NO 39
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the coding strand

<400> SEQUENCE: 39

ttaatggtaa a 11

<210> SEQ ID NO 40
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the coding strand

<400> SEQUENCE: 40

ttaattgtaa a 11

<210> SEQ ID NO 41
<211> LENGTH: 12
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the coding strand

<400> SEQUENCE: 41

ggatttcatt at 12

<210> SEQ ID NO 42
<211> LENGTH: 12
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the coding strand

<400> SEQUENCE: 42

ggattaaatt at 12

<210> SEQ ID NO 43
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the non-coding strand

<400> SEQUENCE: 43

caatagaaga t 11

<210> SEQ ID NO 44
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the non-coding strand

<400> SEQUENCE: 44

caataaaaga t 11

<210> SEQ ID NO 45
<211> LENGTH: 11

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<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the non-coding strand

<400> SEQUENCE: 45

ctttctattg t 11

<210> SEQ ID NO 46
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the non-coding strand

<400> SEQUENCE: 46

ctttccattg t 11

<210> SEQ ID NO 47
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the non-coding strand

<400> SEQUENCE: 47

cccttctcca t 11

<210> SEQ ID NO 48
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the non-coding strand

<400> SEQUENCE: 48

ccctttttcca t 11

<210> SEQ ID NO 49
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the non-coding strand

<400> SEQUENCE: 49

tttaccatta a 11

<210> SEQ ID NO 50
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the non-coding strand

<400> SEQUENCE: 50

tttacaatta a 11

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<210> SEQ ID NO 51
<211> LENGTH: 12
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the non-coding strand

<400> SEQUENCE: 51

ataatgaaat cc 12

<210> SEQ ID NO 52
<211> LENGTH: 12
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide representing the non-coding strand

<400> SEQUENCE: 52

ataatttaat cc 12

<210> SEQ ID NO 53
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: 5'-sequence to the polymorphic sites on the coding strand

<400> SEQUENCE: 53

tacctcccat c 11

<210> SEQ ID NO 54
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: 5'-sequence to the polymorphic sites on the coding strand

<400> SEQUENCE: 54

aaccaaaaac a 11

<210> SEQ ID NO 55
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: 5'-sequence to the polymorphic sites on the coding strand

<400> SEQUENCE: 55

ctgcagtgat g 11

<210> SEQ ID NO 56
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: 5'-sequence to the polymorphic sites on the coding strand

<400> SEQUENCE: 56

taggggttt a 11

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<210> SEQ ID NO 57
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: 5'-sequence to the polymorphic sites on the coding strand

<400> SEQUENCE: 57

atttgaaagg a 11

<210> SEQ ID NO 58
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: 5'-sequence to the polymorphic sites on the non-coding strand

<400> SEQUENCE: 58

gatgggaggt a 11

<210> SEQ ID NO 59
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: 5'-sequence to the polymorphic sites on the non-coding strand

<400> SEQUENCE: 59

tgtttttggt t 11

<210> SEQ ID NO 60
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: 5'-sequence to the polymorphic sites on the non-coding strand

<400> SEQUENCE: 60

catcactgca g 11

<210> SEQ ID NO 61
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: 5'-sequence to the polymorphic sites on the non-coding strand

<400> SEQUENCE: 61

taaaccccct a 11

<210> SEQ ID NO 62
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: 5'-sequence to the polymorphic sites on the non-coding strand

<400> SEQUENCE: 62

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tcctttcaaa t 11

<210> SEQ ID NO 63
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: 3'-sequence to the polymorphic sites on the non-coding strand

<400> SEQUENCE: 63

tgtggatgca a 11

<210> SEQ ID NO 64
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: 3'-sequence to the polymorphic sites on the non-coding strand

<400> SEQUENCE: 64

catggctgct t 11

<210> SEQ ID NO 65
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: 3'-sequence to the polymorphic sites on the non-coding strand

<400> SEQUENCE: 65

agggatctcc c 11

<210> SEQ ID NO 66
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: 3'-sequence to the polymorphic sites on the non-coding strand

<400> SEQUENCE: 66

taaacacctt t 11

<210> SEQ ID NO 67
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: 3'-sequence to the polymorphic sites on the non-coding strand

<400> SEQUENCE: 67

tgttctttat a 11

<210> SEQ ID NO 68
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: 3'-sequence to the polymorphic sites on the coding strand

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<400> SEQUENCE: 68

ttgcatccac a 11

<210> SEQ ID NO 69
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial sequence
<220> FEATURE:
<223> OTHER INFORMATION: 3'-sequence to the polymorphic sites on the coding strand

<400> SEQUENCE: 69

aagcagccat g 11

<210> SEQ ID NO 70
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: 3'-sequence to the polymorphic sites on the coding strand

<400> SEQUENCE: 70

gggagatccc t 11

<210> SEQ ID NO 71
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: 3'-sequence to the polymorphic sites on the coding strand

<400> SEQUENCE: 71

aaaggtgttt a 11

<210> SEQ ID NO 72
<211> LENGTH: 11
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: 3'-sequence to the polymorphic sites on the coding strand

<400> SEQUENCE: 72

tataaagaac a 11

<210> SEQ ID NO 73
<211> LENGTH: 29
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Nuclelotide tail complementary to M13 at its 5'-end.

<400> SEQUENCE: 73

agtcacgacg ttgtaaaacg acggccagt 29

1. An oligonucleotide primer pair suitable for amplifying a polymorphic region of a 5' flanking region of a CYP3A4 gene, wherein the polymorphic region corresponds to position 816 of SEQ ID NO:1.

2. The primer pair of claim 1, having sequences selected from the group consisting of SEQ ID NO:7 and SEQ ID NO:8 and SEQ ID NO:9 and SEQ ID NO:10.

3. A sequence determination oligonucleotide for detecting a polymorphic site in a 5' flanking region of a CYP3A4 gene, said oligonucleotide being complementary to the polymorphic region corresponding to position 461 of SEQ ID NO:1.

4. The oligonucleotide of claim 3, comprising a sequence selected from the group consisting of SEQ ID NO:11; SEQ ID NO:12; SEQ ID NO:13; SEQ ID NO: 14; SEQ ID NO:15; SEQ ID NO:16; SEQ ID NO: 17; and SEQ ID NO:18.

5. A kit comprising at least one oligonucleotide primer pair capable of amplifying the region corresponding to position 461 of SEQ ID NO:1.

6. The kit of claim 5, wherein the primer pair comprises sequences selected from the group consisting of SEQ ID NO: 7 and SEQ ID NO:8 and SEQ ID NO:9 and SEQ ID no: 10.

7. The kit of claim 5, further comprising a sequence determination oligonucleotide complementary to the polymorphic region corresponding to position 461 of SEQ ID NO:1.

8. The kit of claim 7, wherein the oligonucleotide comprises a sequence selected from the group consisting of SEQ ID NO:11; SEQ ID NO:12; SEQ ID NO: 13; SEQ ID NO:14; SEQ ID NO:15;SEQ ID NO:16; SEQ ID NO:17; and SEQ ID NO:18.

9. An oligonucleotide primer pair suitable for amplifying a polymorphic region of a 5' flanking region of a CYP2C9 gene, wherein the polymorphic region corresponds to position 957 of SEQ ID NO:6; position 1049 of SEQ ID NO:6; position 1164 of SEQ ID NO:6; position 1526 of SEQ ID NO:6; position 1661 of SEQ ID NO:6; and position 1662 of SEQ ID NO:6.

10. The primer pair of claim 9, having a sequence selected from the group consisting of SEQ ID NO: 19 and SEQ ID NO:20; SEQ ID NO:21 and SEQ ID NO:22; SEQ ID NO:23 and SEQ ID NO:24; SEQ ID NO:25 and SEQ ID NO:26; SEQ ID NO:27 and SEQ ID NO:28; SEQ ID NO:29 and SEQ ID NO:30; and SEQ ID NO:31 and SEQ ID NO:32.

11. A sequence determination oligonucleotide for detecting a polymorphic site in a 5' flanking region of a CYP2C9 gene, said oligonucleotide comprising a sequence selected from the group consisting of an oligonucleotide complementary to the polymorphic region corresponding to position 957 of SEQ ID NO:6; an oligonucleotide complementary to the polymorphic region corresponding to position 1049 of SEQ ID NO:6; an oligonucleotide complementary to the polymorphic region corresponding to position 1164 of SEQ ID NO:6; an oligonucleotide complementary to the polymorphic region corresponding to position 1526 of SEQ ID NO:6; an oligonucleotide complementary to the polymorphic region corresponding to position 1661 of SEQ ID NO:6; and an oligonucleotide complementary to the polymorphic region corresponding to position 1662 of SEQ ID NO:6.

12. The oligonucleotide of claim 11, comprising a sequence selected from the group consisting of SEQ ID NO:33; SEQ ID NO:34; SEQ ID NO:35; SEQ ID NO:36; SEQ ID NO:37; SEQ ID NO:38; SEQ ID NO:39; SEQ ID NO:40; SEQ ID NO:41; SEQ ID NO:42; SEQ ID NO:43; SEQ ID NO:44; SEQ ID NO:45; SEQ ID NO:46; SEQ ID NO:47; SEQ ID NO:48; SEQ ID NO:49; SEQ ID NO:50; SEQ ID NO:51; SEQ ID NO:52; SEQ ID NO:53; SEQ ID NO:54; SEQ ID NO:55; SEQ ID NO:56; SEQ ID NO:57; SEQ ID NO:58; SEQ ID NO:59; SEQ ID NO:60; SEQ ID NO:61; SEQ ID NO:62; SEQ ID NO:63; SEQ ID NO:64; SEQ ID NO:65; SEQ ID NO:66; SEQ ID NO:67; and SEQ ID NO:68.

13. A kit comprising at least one oligonucleotide primer pair, wherein the primer pair is capable of amplifying a polymorphic region selected from the group consisting of the polymorphic region corresponding to position 957 of SEQ ID NO:6; the polymorphic region corresponding to position 1049 of SEQ ID NO:6; the polymorphic region corresponding to position 1164 of SEQ ID NO:6; the polymorphic region corresponding to position 1526 of SEQ ID NO:6; the polymorphic region corresponding to position 1661 of SEQ ID NO:6; and the polymorphic region corresponding to position 1662 of SEQ ID NO:6.

* * * * *